REMARKS

Claims 1-6, 8, 9, 11-16, 18, 19, 21, 31, 32, 36-58 and 60-65 are pending herein. Claims 3, 8, 9,18, 19, 36, 37, 60 and 61 are canceled herein without prejudice. Claims 1, 4, 5, 12, 38, 40, 42, 44, 46, 48, 50, 52, 54, 56, 62 and 64 are amended herein to more particularly define the invention. Support for these claim amendments is found throughout the specification as set forth below and in the original claim language. It is believed that no new matter is added by these amendments and their entry is respectfully requested. In light of these amendments and the following remarks, applicants respectfully request reconsideration of the pending application and allowance of the pending claims to issue.

Applicants wish to thank Examiner Kaushal and Examiner Priebe for their time and consideration in conducting a personal interview of this case on October 16, 2002 at the U.S. Patent and Trademark Office, at which Dr. Dominique Robertson, Dr. Karen Magri and Dr. Mary Miller were in attendance. The pending enablement and written description rejections were discussed and claim amendments were proposed which applicants believe should place the pending claims in condition for allowance. These amendments are presented herein and described below, along with other issues discussed during the interview.

I. Rejection under 35 U.S.C. § 112, first paragraph

Claims 1-6, 8-9, 11-16, 18-19, 21, 31-32, 36-58 and 60-65 remain rejected as allegedly containing subject matter which was not described in the specification in such a way as to reasonably convey to one skilled in the art that the inventor, at the time the application was filed, had possession of the claimed invention for reasons of record. The Office Action states that previous arguments provided by the applicant were found to be unpersuasive and cites various case law in support of the contention that one skilled in the art would conclude that applicant was not in possession of the

claimed genus because a description of only one member of the genus is not representative of the variants of the genus and is insufficient to support the claim.

As discussed during the personal interview on October 16, 2002 with Examiners Kaushal and Priebe, applicants believe that the specification as filed provides adequate written description of the claimed invention and that this rejection should be withdrawn. In particular, it is acknowledged in the Office Action on page 4 and stated in the Guidelines for Written Description that possession may be shown by an actual reduction to practice of an invention. Applicants have reduced to practice a geminvirus silencing vector comprising a geminivirus genome comprising the geminivirus AL1, AL2 and Al3 coding sequences and comprising an heterologous DNA comprising at least a fragment of a gene endogenous to a plant and having all of the additional elements as set forth in claim 1 of the present invention. Thus, applicants have met the requirements for written description of the claimed invention and applicants further note that both Examiners Kaushal and Priebe concurred with this conclusion during the personal interview. Therefore, applicants believe this rejection has been overcome and respectfully request its withdrawal and allowance of the pending claims to issue.

II. Rejection under 35 U.S.C. § 112, first paragraph

Claims 1-6, 8-9, 11-16, 18-19, 21, 31-32 and 35-65 stand rejected under 35 U.S.C. §112, first paragraph, as allegedly being enabling only for the silencing of the magnesium cheletase (su) gene and the luciferase (luc) gene in *N. benthamiana* using a TGMV vector, but not for any and all geminvirus silencing vectors comprising any and all geminivirus genomes, a DNA construct comprising any and all geminivirus silencing vectors, wherein the genome contains heterologous DNA which has at least 80-95% sequence similarity to any and all genes endogenous to any and all plants. The Office Action further states that applicant's arguments have been considered but not found persuasive and that the amount of experimentation required would include making and testing any and all gene sequences from any and all plant species that have at least 80-95% sequence homology to any and all genes endogenous to any and

all species of plant. The Office Action goes on to state that the experimentation required would further include the use of DNA variants (as claimed) in silencing of any and all endogenous genes of any and all species of plants. For these reasons, it is the Examiner's conclusion that the claims are not enabled commensurate with their scope.

Applicants respectfully traverse this rejection and assert that the invention as claimed is adequately enabled for the full scope of the pending claims. Specifically, as discussed at the personal interview with Examiners Kaushal and Priebe on October 16, 2002, applicants have demonstrated that the claimed silencing vectors function as claimed in studies employing a variety of geminivirus genomes, a variety of heterologous DNA sequences and a variety of plants. Applicants provide herein evidence of two different geminivirus vectors, 17 different heterologous DNA sequences and species from two different plant families in which the claimed invention has been demonstrated to function as described in the form of a Declaration of Dr. Dominique Robertson under 37 C.F.R. § 1.132, attached hereto as Appendix B.

Furthermore, claims 1, 12, 38, 40, 42, 44, 46, 48, 50, 52, 54, 56, 62 and 64 are amended herein to recite a heterologous DNA comprising at least a fragment of a gene endogenous to a plant rather than a heterologous DNA having at least 80% sequence similarity to a gene endogenous to a plant. This amendment was discussed with Examiner Kaushal and Examiner Priebe, who agreed that the amendatory language provided more clarity to the claims. Support for this amendment can be found throughout the specification, and in particular, for example, on page 3, lines 1-3, lines 6-9 and lines 11-13; page 5, line 28 through page 6, line 1; page 9, lines 17-19; page 10, line 26; page 11, lines 7-13; and page 12, lines 27-30. Support can also be found in the Examples provided in the specification, wherein several vectors are described which comprise a fragment of an endogenous plant gene.

Claims 4 and 5 are amended herein at the suggestion of Examiner Priebe, to be consistent with the language of claim 1, from which these claims depend. Support for

the amendments to claims 4 and 5 is found in the language of claim 3 and throughout the specification.

Claim 64 is amended herein for clarity to recite that a nucleic acid sequence is introduced into said plant cell, rather than provided to said plant cell. Support for this amendment can be found throughout the specification and particularly in the Examples provided, wherein the introduction of nucleic acid into plant cells is described.

For the reasons provided above, applicants believe that the invention is fully enabled commensurate with the claims and respectfully request that this rejection be withdrawn.

The Examiner is encouraged to contact the undersigned directly if such contact will expedite the examination and allowance of the pending claims.

A check in the amount of $ 200.00 is enclosed in payment of the fee for a two month extension of time. This amount is believed to be correct. However, the commissioner is hereby authorized to charge any deficiency or credit any overpayment to Deposit Account No. 50-0220.

Respectfully submitted,

Mary L. Miller
Registration No. 39,303

CERTIFICATE OF MAILING
I hereby certify that this correspondence is being deposited with the United States Postal Service as first class mail in an envelope addressed to: Commissioner For Patents, Washington, DC 20231, on November 26, 2002.

Clara R. Beard

APPENDIX A

Marked-up Version Showing Changes Made

1. (Thrice Amended) A geminivirus silencing vector comprising a geminivirus genome comprising:

the geminivirus AL1, AL2 and AL3 coding sequences,

heterologous DNA, said heterologous DNA [having at least 80% sequence similarity to] comprising at least a fragment of a gene endogenous to a plant that occurs naturally in the plant genome,

wherein said heterologous DNA is constitutively expressed and said AL1, AL2 and AL3 coding sequences are bidirectionally transcribed from said geminivirus silencing vector, and

wherein said geminivirus silencing vector silences expression of the endogenous plant gene upon introduction into a plant cell.

4. (Twice Amended) A vector according to claim [3] 1, wherein said heterologous DNA is operably associated with a promoter [is the promoter] that is associated with said endogenous plant gene.

5. (Twice Amended) A vector according to claim [3] 1, wherein said heterologous DNA is operably associated with [promoter is] the geminivirus coat protein promoter.

12. (Thrice Amended) A DNA construct comprising a geminivirus genome, wherein the DNA encoding the geminivirus coat protein has been replaced in part or in total with heterologous DNA [having at least 80% sequence similarity to] comprising at least a fragment of an endogenous plant gene that occurs naturally in the plant genome.

38. (Twice Amended) A geminivirus silencing vector comprising a geminivirus genome which contains a heterologous DNA, said heterologous DNA [having at least 80% sequence similarity to] comprising at least a fragment of a coding region of a gene endogenous to a plant, wherein the heterologous DNA sequence is inserted into the silencing vector in the antisense orientation, and wherein said geminivirus silencing vector silences expression of the endogenous plant gene upon introduction into a plant cell.

40. (Twice Amended) A DNA construct comprising a geminivirus genome, wherein the DNA encoding the geminivirus coat protein has been replaced in part or in total with heterologous DNA [having at least 80% sequence similarity to] comprising at least a fragment of a coding region of a gene endogenous to a plant, and wherein the heterologous DNA sequence is inserted into the geminivirus genome in the antisense orientation.

42. (Twice Amended) A geminivirus silencing vector comprising a Tomato Golden Mosaic Virus (TGMV) genome which contains heterologous DNA, said heterologous DNA [having at least 80% sequence similarity to] comprising at least a fragment of a gene endogenous to a plant, wherein said geminivirus silencing vector silences expression of the endogenous plant gene upon introduction into a plant cell.

44. (Twice Amended) A geminivirus silencing vector comprising an African Cassava Mosaic Virus (ACMV) genome which contains heterologous DNA, said heterologous DNA [having at least 80% sequence similarity to] comprising at least a fragment of a gene endogenous to a plant, and wherein said geminivirus silencing vector silences expression of the endogenous plant gene upon introduction into a plant cell.

46. (Twice Amended) A DNA construct comprising a Tomato Golden Mosaic Virus (TGMV) genome, wherein the DNA encoding the TGMV coat protein

has been replaced in part or in total with heterologous DNA [having at least 80% sequence similarity to] comprising at least a fragment of an endogenous plant gene.

48. (Twice Amended) A DNA construct comprising an African Cassava Mosaic Virus (ACMV) genome, wherein the DNA encoding the ACMV coat protein has been replaced in part or in total with heterologous DNA [having at least 80% sequence similarity to] comprising at least a fragment of an endogenous plant gene.

50. (Twice Amended) A method of silencing the expression of an endogenous plant gene in a plant cell, comprising inoculating said plant cell with a geminivirus silencing vector comprising a geminivirus genome which contains heterologous DNA, said heterologous DNA [having at least 80% sequence similarity to] comprising at least a fragment of a gene endogenous to a plant.

52. (Twice Amended) A method of silencing the expression of an endogenous plant gene in a plant cell, comprising inoculating said plant cell with a DNA construct comprising a geminivirus genome, wherein the DNA encoding the geminivirus coat protein has been replaced in part or in total with heterologous DNA [having at least 80% sequence similarity to] comprising at least a fragment of an endogenous plant gene.

54. (Twice Amended) A method of systemically silencing expression of an endogenous plant gene in a plant, comprising inoculating said plant with a geminivirus silencing vector comprising a geminivirus genome which contains heterologous DNA, said heterologous DNA [having at least 80% sequence similarity to] comprising at least a fragment of a gene endogenous to a plant.

56. (Twice Amended) A method of systemically silencing expression of an endogenous plant gene in a plant, comprising inoculating said plant with a DNA construct comprising a geminivirus genome, wherein the DNA encoding the

geminivirus coat protein has been replaced in part or in total with heterologous DNA [having at least 80% sequence similarity to] comprising at least a fragment of an endogenous plant gene

62. (Amended) A geminivirus silencing vector comprising a Tomato Golden Mosaic Virus (TGMV) genome comprising:

the TGMV AL1, AL2 and AL3 coding sequences operably associated with an AL1 promoter,

heterologous DNA, said heterologous DNA operably associated with a TGMV coat protein promoter and [having at least 80% sequence similarity to] comprising at least a fragment of a gene endogenous to a plant that occurs naturally in the plant genome,

wherein said heterologous DNA and said AL1, AL2 and AL3 coding sequences are bidirectionally transcribed from said geminivirus silencing vector, and

wherein said geminivirus silencing vector silences expression of the endogenous plant gene upon introduction into a plant cell.

64. (Amended) A method of silencing the expression of an endogenous plant gene in a plant cell, comprising:

[providing] introducing a nucleic acid sequence encoding the geminivirus movement proteins [to] into said plant cell;

inoculating said plant cell with a geminivirus silencing vector comprising a geminivirus genome which contains heterologous DNA [having at least 80% sequence similarity to] comprising at least a fragment of a gene endogenous to a plant.

Exhibit A

A. 1 T mato G ld n Mosaic Virus V ctors, as describ d in P el t al. (2001) Plant J. 27: 357

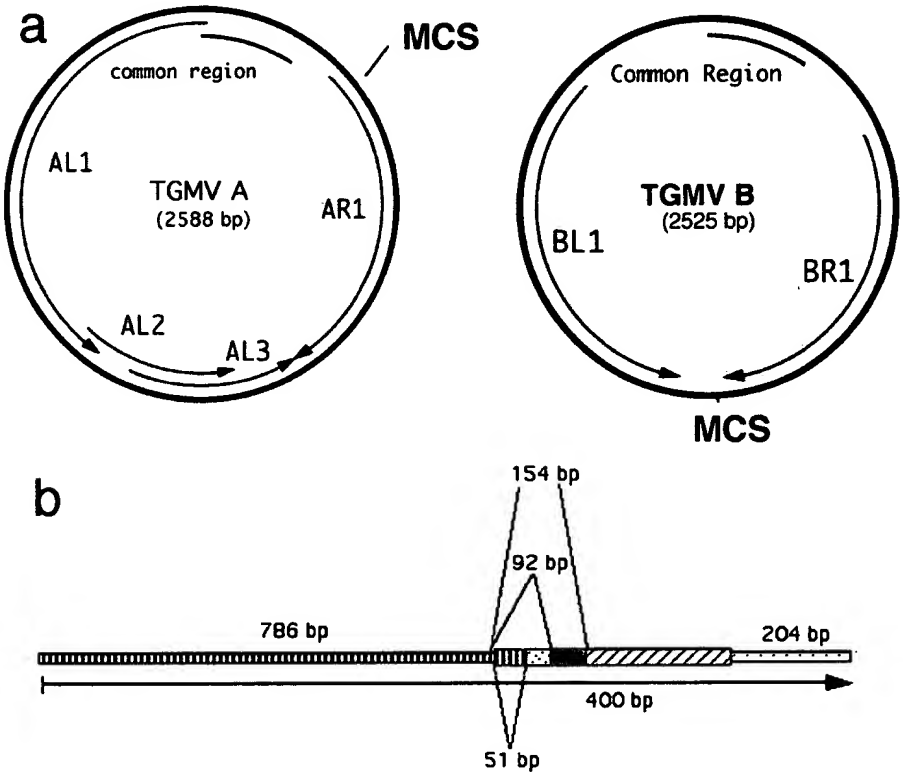


Figure 1. TGMV A- and B-derived episomal silencing vectors.
(a) Silencing DNA fragments homologous to endogenous gene(s) are inserted into a multiple cloning site (MCS). In the A component vector, silencing fragments are transcribed from the AR1 promoter and are inserted in place of the AR1 gene. In the B component vector, silencing DNA is inserted 20 bp downstream of the BR1 stop codon, and is cotranscribed with BR1. The viral genes, AL1, AL2, AL3, BL1 and BR1, are needed for replication and movement of the vector. The common region is identical in the two components and contains the origin of replication.
(b) Location of gene fragments from the su cDNA used for silencing. The arrow shows the su cDNA and the ATG and TAA mark the beginning and end of the gene.

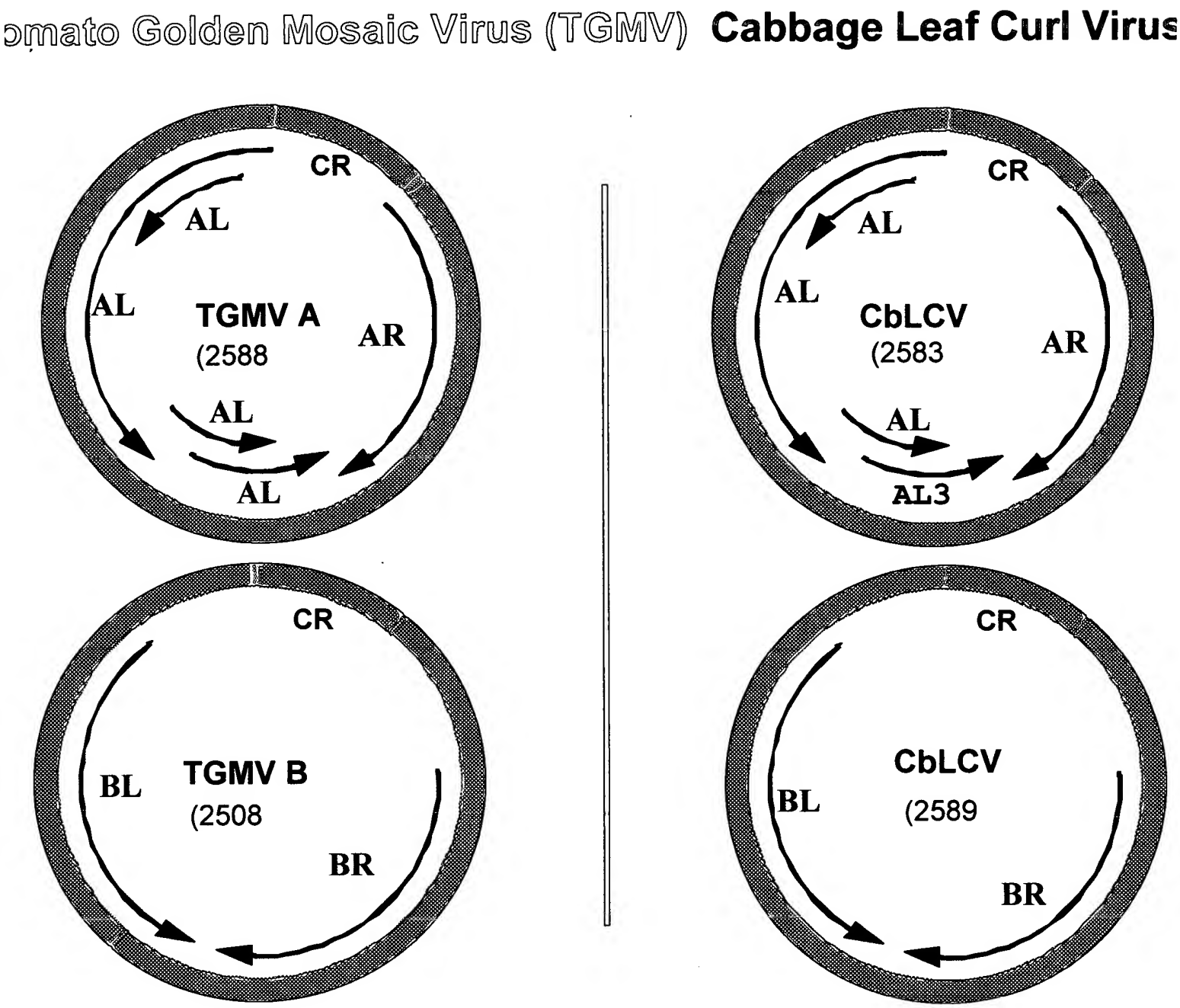
The TGMV A vector is a replacement vector. Heterologous DNA replaces the AR1 gene and is transcribed using the AR1 promoter. The TGMV B is an insertional vector. Heterologous DNA is inserted into the MCS \ and is cotranscribed with the BR1 gene. The size limitation is approximately 160 bp. This work proves that geminiviruses with

monopartite genomes, having no dispensable genes such as AR1, can be engineered as insertional vectors for gene silencing.

A.2. Consensus homology between N. benthamiana (endogenous gene) and N. tabacum (silencing fragment) for magnesium chelatase. In Figure 1b, section A.1., a Nicotiana tabacum cDNA sequence for a gene encoding one of three subunits of the enzyme magnesium chelatase (required for chlorophyll formation) is shown. Below is a comparison of the sequences in N. benthamiana (target gene for silencing) and N. tabacum (heterologous sequences cloned into TGMV to initiate silencing in N. Benthamiana). Fragments of the N. tabacum cDNA between 91 and 795 bp produced reliable silencing. This demonstrates that 100% homology between the target and the viral vector insert (heterlogous DNA) is not absolutely required for silencing to occur. N indicates a non-conserved nucleotide.

ATGGCTTCACTnnTnGGnACTTCCTCTTCAGCAGCAGCTGCTGCAATATT
AGCTTCTACACCCTTnTCTTCTCGCTCCTnTAAnnCTnCCnTTTTCTCCC
TCTTCCCTTCTTCAGGGCAGnGTCAAGGGAGGAAGTTTTATGGAGGGATT
AGAnTCCCAGTTAAGAAAGGGAGGTCCCAATTnCATGTGGCAATTTCAAA
TGTTGCnACGGAAnTCAACCCTGCTCAAGAACAGGGTCAGAAACTTGCTG
AGGAGAGCCAGAGACCnGTGTATCCATTTGCAGCTATAGTGGGACAAGAn
GAnATGAAGTTATGTCTTTTGCTGAATGTAATTGATCCAAAGATTGGAGG
TGTGATGATAATGGGTGATnGnGGnACCGGGAAGTCCACCACGGTTAGAT
CTTTGGTAGATTTACTTCCTGAnATCAAAGTTATTTCTGGTGATCCGTTC
AATTCAGATCCAGATGACCAAGAAGTAATGAGTGCAGAAGTCCGTGACAA
ATTGAGGAGCGGAnAGnAGCTTCCTATATCTCGTACCAAAATCAACATGG
TTGATTTACCGCTAGGTGCTACTGAnGACAGGGTGTGTGGCACAATCGAC
ATTGAGAAAGCTCTTACTGAGGGTGTGAAGGCTTTCGAnCCTGGTCTTCT
TGCTAAAGCTAACAGAGGAATnCTTTATGTCGATGAnGTTAATCTTTTGG
AnGACCATTTAGTAnATGTTCTTTTGGATTCTGCAGCATCGGGATGGAAC
ACTGTTGAAAGAnAnGGGATATCAATnTCACAnCCnGCCCGATTTATCCT
TATTGGTTCnGGTAATCCTGAAnAAnGAGAACTTAnGCCACAACTTCTTG
ATCGATTTGGAATGCATGCCCAAGTGGGGACCGTGAGAGATGCAGAGCTG
AGAGTGAAGATCGTTGAGGAAAGAGCTCGTTTTGATAAGAACCCCAAGGA
ATTCCGnGAGTCATACAAGGCAGAGCAAGAAAAGCTCCAGAATCAAATCG
ACTCAGCTAGGAACGCTCTTTCTGCTGTTACAATnGATCATGATCTTCGA
GTTAAAAnCTCTAAGGTCTGTGCAGAACTnAAnGTCGATGGATTGAGAGG
TGATATAGTCACTAACAGGGCAGCAnGAGCGTTGGCTGCACTAAAAGGAA
GAGATAAGGTnACTCCGGAnGATATCGCCACTGTCATTCCCAACTGCTTA
AGACACAGnCTnAGnAAnGAnCCnT

A.3. Comparison of cabbage leaf curl virus and tomato golden mosaic virus genomes. The genus begomovirus has a conserved genome structure.



A.4. Two diff rent viruse carrying DNA fragm nts that targ t homologous gen s (coding for a subunit of magne ium chelata e) required for chlorophyll formation cau eff ctiv sil ncing and lo s of chlorophyll in both N. benthamiana and Arabidopsis.

Fig. 2. TGMV carrying a 154 bp DNA fragment (SU) inoculated into N. benthamiana.

Fig. 3. CbLCV carrying a 400 bp DNA fragment (CH42) inoculated into Arabidopsis.

A.5. Genome conservation in members of the Geminiviridii, genus begomovirus.

Table 1 lists the amino acid similarity of selected geminiviruses and demonstrates that cabbage leaf curl virus, with only 69% amino acid identity with tomato golden mosaic virus, silences as effectively as tomato golden mosaic virus. Table from Turnage et al. (2002) The Plant J. 30:107. Work in another lab (Atkinson et al. 1998 The Plant J. 15:593) demonstrates the monopartite geminiviruses of the genus mastrevirus (tobacco yellow dwarf virus) can also be engineered for silencing vectors. They developed an insertional vector for silencing chalcone synthase in Petunia.

Table 1. Amino acid similarity of selected new world (NW) and old world (OW) begomoviruses

Virus	Average[a]	AL1/AC1[a]	BL1/BC1[a]
Cabbage leaf curl virus	**100 (100)**	**100 (100)**	**100 (100)**
Bean calico mosaic virus	76 (86)	84 (90)	75 (87)
Squash leaf curl virus	74 (84)	81 (88)	75 (86)
Sida golden mosaic virus	73 (84)	63 (76)	82 (90)
Cucurbit leaf crumple geminivirus	73 (84)	79 (88)	74 (86)
Bean golden mosaic virus	72 (84)	63 (75)	82 (91)
Dicliptera yellow mottle virus	72 (82)	63 (74)	82 (89)
Bean dwarf mosaic virus	72 (82)	63 (75)	82 (89)
Tomato mottle virus	71 (82)	63 (76)	80 (89)
Tomato leaf curl virus	71 (81)	65 (75)	81 (89)
Tomato leaf crumple virus	71 (83)	65 (78)	80 (89)
Chino del tomato virus	71 (83)	64 (77)	81 (89)
Potato yellow mosaic virus	70 (80)	63 (75)	82 (88)
Taino tomato mottle virus	70 (82)	62 (76)	80 (90)
Abutilon mosaic virus	69 (81)	59 (73)	81 (89)
Tomato golden mosaic virus	**69 (81)**	**60 (74)**	**78 (88)**
Tomato rugose mosaic virus	68 (80)	60 (74)	76 (86)
Pepper huasteco virus	67 (80)	52 (68)	82 (91)
Havana tomato virus	66 (78)	62 (75)	70 (79)
Cassava latent virus	44 (60)	53 (68)	44 (61)
Indian cassava mosaic virus	42 (60)	51 (68)	43 (58)
Vigna mungo yellow mosaic virus	42 (61)	52 (68)	44 (63)
South African cassava mosaic virus	41 (58)	51 (67)	42 (59)
Mungbean yellow mosaic virus	41 (59)	51 (67)	43 (61)
Watermelon chlorotic stunt virus	41 (59)	51 (68)	43 (58)
West African cassava mosaic virus	40 (57)	51 (67)	41 (56)
Indian mungbean yellow mosaic virus	40 (60)	51 (67)	44 (62)
Tomato yellow leaf curl virus	39 (59)	53 (68)	39 (59)

Exhibit B Summary of genes that have been silenced using geminiviruses

B.1. Nicotiana benthamiana inoculated with TGMV vectors carrying different inserts.

Construct	Heterologous fragment	Target gene	Silencing phenotype
pSK16L	Luciferase, 650 bp	Accession no. P08659	Loss of transgenic luciferase luminescence (Kjemtup et al., 1998)
pCPTGMV::GFP	Green fluorescent protein, 780 bp	Accession no. AAB47998	Loss of GFP fluorescence (Peele et al., 2001)
pCPTGMV A::su	786 bp *Acc*651/*Eco*RV fragment, corresponding to nt 0-786 of su cDNA, antisense orientation	Accession no. AAG35472	Loss of chlorophyll (Kjemtrup et al., 1998; Peele et al., 2001)
pCP1.3BPCNA	Proliferating cell nuclear antigen, 122 bp	Accession No. AF486816	Loss of primary plant growth (Peele et al., 2001)
pNMTGMV B::Clone 9	106 bp *Ssp*I/*Eco*RV fragment, corresponding to 96 bp 5' cDNA of the Clone 9 961 bp fragment upregulated by viral replication	Unpublished, homology to (*A. thaliana*) NP563717.1	Loss of viral symptoms in new growth (Muangsan, Ph.D. thesis 2002)
pNMTGMV B::Clone 37	Unknown protein (clone 37) upregulated by viral replication protein	Unpublished, homology to (*A. thaliana*) AAD27878	Loss of viral symptoms and DNA in new growth (Eagle, Muangsan and Robertson, unpublished)
pNMTGMV B::Clone 8	Unknown proteins (clone 8) upregulated by viral replication	Unpublished	Loss of viral symptoms in new growth (Eagle, Muangsan and Robertson, unpublished)
pNMTGMV B::Clone 25.1	Clone 25.1, with homology to calmodulin-related protein	Unpublished, homology to (*A. thaliana*) NP198593	Loss of viral symptoms in new growth (Eagle, Muangsan and Robertson, unpublished)
pCJTGMV::Rb	150 bp fragment of tobacco Retinoblastoma related protein	BAA76477	Severe necrosis in inoculated leaf, lesions in upper leaves, attenuated symptoms, flowers curl (Jordan and Robertson, unpublished)

B.2. Phenotypic changes reflecting downregulation of target genes by TGMV silencing vectors

Figure 1. Attenuated symptoms in *Nicotiana benthamiana* plants silenced in *clone 9*. Plants were inoculated with a combination of TGMV A and TGMV B::SU (d) or TGMV A and TGMV B::Clone 9 (e) and photographed four weeks post inoculation. Arrow (e) show symptoms in lower leaves that were eliminated in new growth.

Figure 2. N. benthamiana silenced with TGMV carrying a DNA fragment from clone 37. Mild symptoms are shown (arrow) in newly infected tissue but upper growth shows no sign of symptoms and lacks viral DNA.

Fig. 3 N. benthamiana inoculated with TGMV vector containing a fragment of the retinoblastoma gene. Left, cell death in vegetative growth and attenuated viral symptoms. Right, flower curling 360°. Normally, flowers are long and straight.

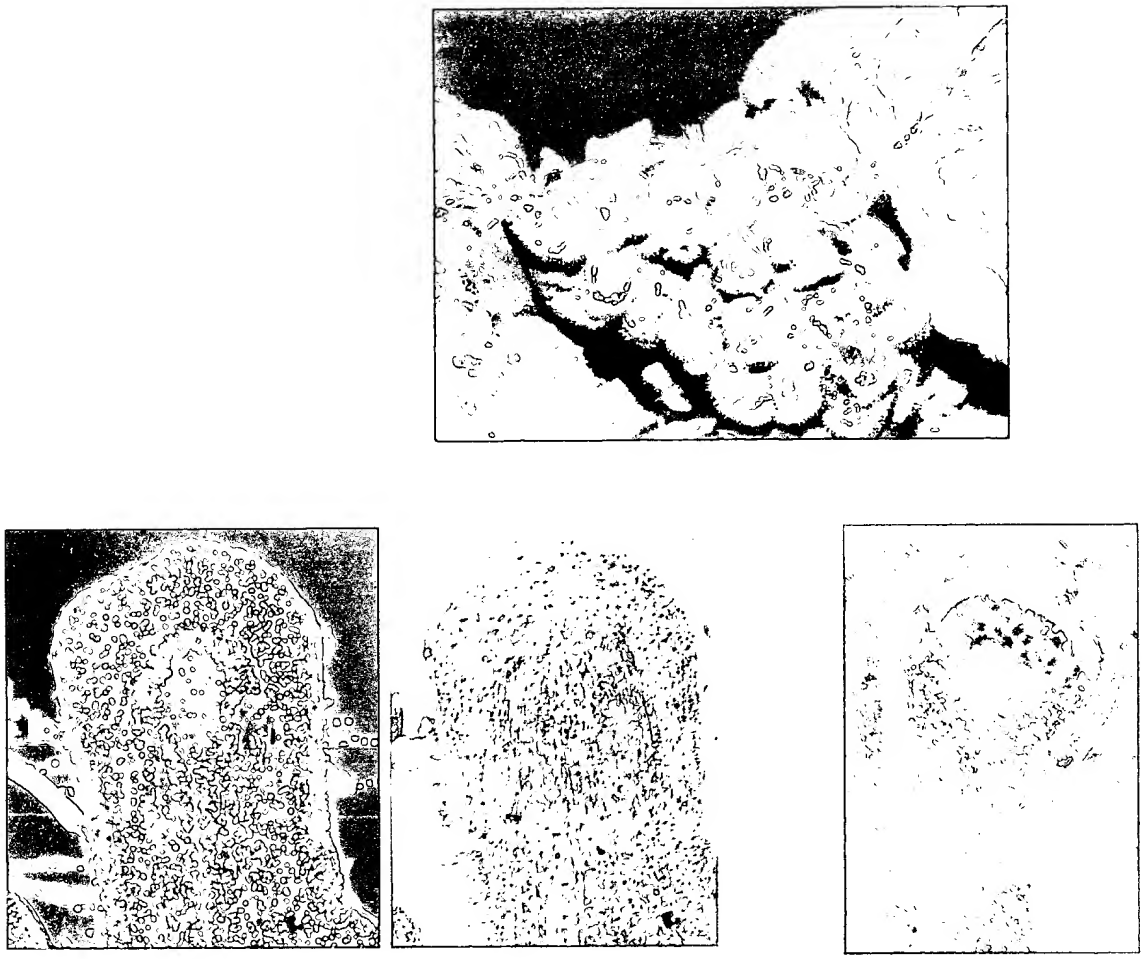


Fig. 4 N. benthamiana silenced for PCNA. Top – loss of apical growth. Bottom left, immunolocalization of PCNA in silenced meristems (center) demonstrates loss of PCNA expression compared to wt (right). Left shows the same meristems as center visualized for nuclei using DAPI.

B.3. Cabbage Leaf Curl Vect rs inoculated into Arabidopsis

CbLCV construct	Heterologous DNA	gene	phenotype
pMTCbLCVA::CH42A	360 bp of homology to the *ch42* gene, antisense orientation	accession no. P16127	Loss of chlorohyll (Turnage et al., 2002)
pNMCbLCVA::CH42S	406 bp fragment with 374 bp of homology to the *ch42* gene, sense orientation	accession no. P16127	Loss of chlorohyll (Turnage et al., 2002)
pNMCbLCVA::luc	618 bp of homology to the *luciferase* gene, sense orientation	Accession no. P08659	Control, no homology to endogenous gene
pNMCbLCVA::PCNA	412 bp fragment, corresponding to nt 115-526 of *PCNA1* cDNA, antisense	accession No. NM100611	Reduced viral accumulation
pNMCbLCVA::CaMRP	400 bp fragment, corresponding to nt 21-420 of *CaMRP c*DNA, antisense	accession No. AY117325	No detectable phenotype
pNMCbLCVA::AtG1RP	398 bp fragment, corresponding to nt 1026 to 1421 of *AtG1RP* cDNA, antisense	accession No. AY054498	No detectable phenotype
pNMCbLCVA::SGS2	434 bp fragment, corresponding to nt 2211-2644 of *SGS2* cDNA, antisense	Accession No. AF239718	Reduces CH42 silencing (light green vs white)
pNMCbLCVA::SDE3	437 bp fragment, corresponding to nt 1591-2027 of *SDE3* cDNA, antisense	accession no. AF339908	CH42 silencing is uneven
pMTCbLCVA::PDS	370 bp fragment from phytoene desaturase	Accession no. AAA20109	Loss of chlorophyll and carotenoids (Turnage et al., 2002)
pMTCbLCVA::GFP	388 bp fragment from mGFP5	Accession no. AAB47998	Loss of green fluorescence in GFP transgenic plants (Turnage et al., 2002)
pNMCbLCVB::CH42	144 bp *Bam*HI/*Eco*RV fragment of the	accession no. P16127	Loss of chlorohyll (Turnage et al.,

	ch42 cDNA, antisense orientation		2002)
pNMCbLCVB::CH42	144 bp *Bam*HI/*Eco*RV fragment of the *ch42* cDNA, sense	accession no. P16127	Loss of chlorohyll (Turnage et al., 2002)

B.4. Selected examples of silencing phenotypes in Arabidopsis inoculated with CbLCV vectors carrying different inserts

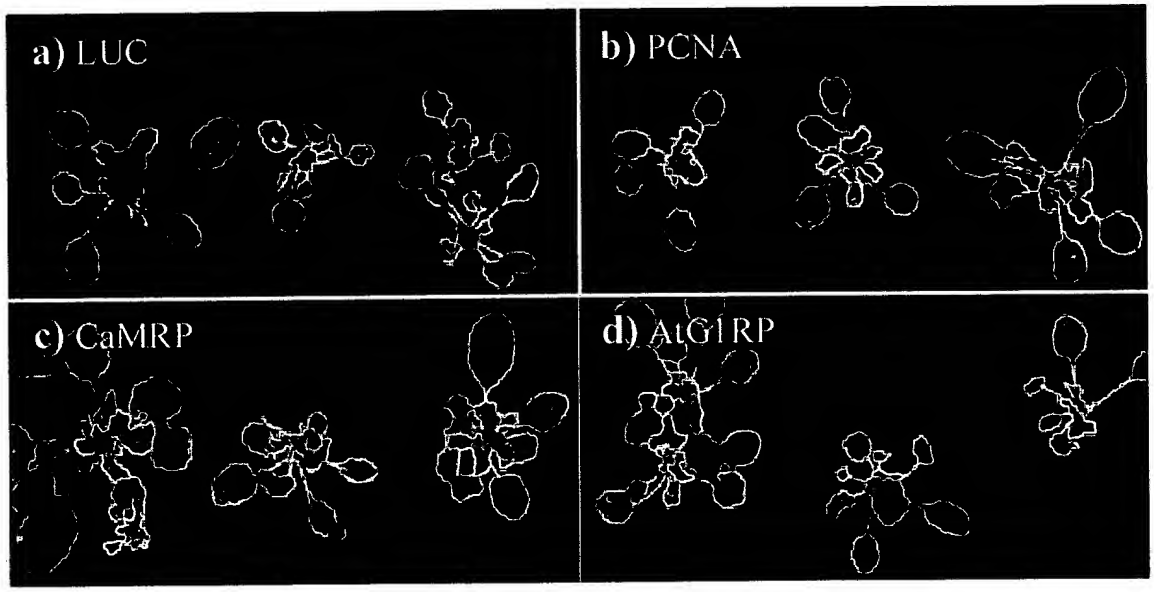
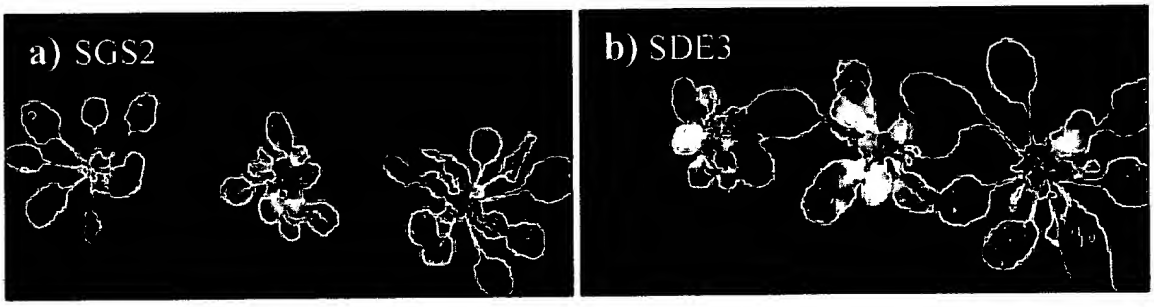


Figure 7. VIGS of *PCNA, CaMRP*, and *AtG1RP* endogenous genes.
Wild type Arabidopsis plants four-weeks-old were inoculated with CbLCV::luc or CbLCV carrying a fusion of *CH42* fragment and one of these three genes: *PCNA, CaMRP,* or *AtG1RP*. CH42 silencing, yellow tissue, were observed in PCNA (b), CaMRP (c) and AtG1RP (d) plants. Photographs were taken at 25 dpi.



A.5. Genom cons rvation in memb rs of the Geminiviridii, genus b g movirus.

Table 1 lists the amino acid similarity of selected geminiviruses and demonstrates that cabbage leaf curl virus, with only 69% amino acid identity with tomato golden mosaic virus, silences as effectively as tomato golden mosaic virus. Table from Turnage et al. (2002) The Plant J. 30:107. Work in another lab (Atkinson et al. 1998 The Plant J. 15:593) demonstrates the monopartite geminiviruses of the genus mastrevirus (tobacco yellow dwarf virus) can also be engineered for silencing vectors. They developed an insertional vector for silencing chalcone synthase in Petunia.

abl 1. Amino acid similarity of selected new world (NW) and old world (OW) begomoviruses

/irus	Average[a]	AL1/AC1[a]	BL1/BC1
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3ean calico mosaic virus	76 (86)	84 (90)	75 (87)
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' **mat golden mosaic virus**	**69 (81)**	**60 (74)**	**78 (88)**
'omato rugose mosaic virus	68 (80)	60 (74)	76 (86)
'epper huasteco virus	67 (80)	52 (68)	82 (91)
lavana tomato virus	66 (78)	62 (75)	70 (79)
:assava latent virus	44 (60)	53 (68)	44 (61)
ndian cassava mosaic virus	42 (60)	51 (68)	43 (58)
/igna mungo yellow mosaic virus	42 (61)	52 (68)	44 (63)
5outh African cassava mosaic virus	41 (58)	51 (67)	42 (59)
/lungbean yellow mosaic virus	41 (59)	51 (67)	43 (61)
Vatermelon chlorotic stunt virus	41 (59)	51 (68)	43 (58)
Vest African cassava mosaic virus	40 (57)	51 (67)	41 (56)
ndian mungbean yellow mosaic virus	40 (60)	51 (67)	44 (62)